MCH Released in a Novel Pheromone Dispenser Prevents Spruce Beetle, *Dendroctonus rufipennis* (Coleoptera: Scolytidae), Attacks in South-central Alaska

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ABSTRACT Field tests of 3-methyl-2-cyclohexen-1-one (MCH), the antiaggregation pheromone of the spruce beetle, *Dendroctonus rufipennis* Kirby, were conducted in south-central Alaska in stands of Lutz spruce, *Picea* x *lutzii* Little, and Sitka spruce, *P. sitchensis* (Bong.) Carr. The deployment of MCH in a novel releaser significantly reduced trap catches and spruce beetle attacks on standing live spruce by 96 and 87%, respectively. The results of this study demonstrate the first successful field test of MCH in Alaska for the prevention of spruce beetle attacks on standing, live spruce.

KEY WORDS Bark beetles, *Dendroctonus rufipennis*, semiochemicals, antiaggregation, *Picea* sp., AK (south-central)

THE SPRUCE BEETLE, *Dendroctonus rufipennis* (Kirby), is the most significant natural mortality agent of mature spruce in the United States (Holsten et al. 1999). At the peak of an on-going outbreak in Alaska, >500,000 ha of spruce stands were infested in 1 yr with ≈30 million trees being killed annually (Wittwer 2000).

Silvicultural treatments, such as thinning, that maintain more resistant stands with moderate growth of residual trees are important tactics for reducing the susceptibility of spruce stands to spruce beetle outbreaks (Sartwell and Stevens 1975, Hard and Holsten 1985, Holsten et al. 1999). For areas in which silvicultural manipulations are neither desirable nor possible, other tactics for reducing damage to spruce beetle, such as aggregant and antiaggregant pheromones, are being developed (Holsten 1994, Werner and Holsten 1995). MCH (3-methyl-2-cyclohexen-1-one) has been shown to inhibit aggregation of the spruce and Douglas-fir beetle (*D. pseudotsugae* Hopkins) (Kline et al. 1974, Furniss et al. 1974, 1977, McGregor et al. 1984). In a long-term search for an effective release device for MCH, numerous devices have been field tested. Examples include: 1) glass vials with liquid MCH with a release rate per vial of 0.5–0.9 mg per hr (Rudinsky et al. 1972); 2) granular formulations releasing from 2 to 10 mg per day per ha depending on application rate (Holsten and Werner 1984); and 3) bubble caps releasing from 4–5 mg per day per bubble cap with ≈75 bubble caps per ha used (Holsten and Werner 1987, Lindgren et al. 1989, Ross et al. 1996).

There have, however, been a number of successful applications of antiaggregation pheromones for the management of forest pests. Aerial applications of verbenone in Northwestern Montana significantly reduced the attack of lodgepole pine, *Pinus contorta* variety *latifolia* Engelmann, by the mountain pine beetle, *D. ponderosae* Hopkins, in one of 2 yrs (Shea et al. 1992). A ground application of MCH released from bubble caps was effective in reducing the probability of Douglas-fir beetle infestations occurring in high risk stands in Washington and Idaho (Ross and Daterman 1995). There have also been, however, failures with the use of antiaggregation pheromones to reduce tree attack by the mountain pine beetle (Amman 1993, Borden 1995) and the spruce beetle (Holsten and Werner 1985, 1987, Zogas 2001). These failures could be caused by technical problems involving the deployment, consistent release (Holsten et al. 2001), longevity of releasers, and dispersion of semiochemicals from the releaser, as well as bark beetle population densities and the influence of microclimate within the stand.

The characteristics and features of an ideal pheromone release system (Jutsum and Gordon 1989) include: 1) release a constant amount of pheromone per unit of time, independent of temperature, humidity, and light; 2) have the ability to release different pheromones; 3) have the ability to provide different release rates; 4) protect the pheromone from degradation; 4) release all the pheromone; and 5) release the

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pheromone for a short or long time, depending on the pest/crop. Commonly used releasers such as bubble caps and beads are first-order emitters, which means that the rate of pheromone release decreases with time. This is an unwanted behavior, because pheromone plume concentration thresholds have to be maintained for effective treatment during the beetle flight period. Med-e-Cell (San Diego, CA) has developed a disposable microinfusion pump, currently used for drug delivery that has been modified to precisely deliver, continuously, a metered amount of semiochemical into the environment. Because this zero-order releaser is not diffusion-dependent, it is considerably less temperature-sensitive than other semiochemical release devices.

Our objectives were: 1) to compare the Med-e-Cell device to the commonly used bubble cap dispenser for reducing spruce beetle catch in attractive traps and 2) to evaluate the efficacy of the Med-e-Cell device for protecting live spruce trees.

Materials and Methods

Study Site Characteristics. In 1999, traps were placed among Lutz spruce, *Picea* x *lutzii* Little, trees at 300 m elevation in the Hillside area of Anchorage, AK. This predominately Lutz spruce stand contained trees that were ≈90 yr old with a mean diameter at breast height (dbh) (dbh @ 1.3 m) of 15 cm, a mean height of 17 m, and a stand density of ≈1250 stems per ha. Less than 10% of the stand contained mature paper birch, *Betula papyrifera* Marshall. Shrub cover was sparse, consisting mostly of labrador tea, *Ledum groenlandicum* L., and clubmoss, *Lycopdium* spp. Spruce beetle activity in the study site was declining with ≈28% of the stand dead because of beetle attack.

In 2000, MCH release devices were placed among mature Sitka spruce, *P. sitchensis* (Bong.) Carr., at 250 m elevation near the confluence of the Snow River and Kenai Lake on the Kenai Peninsula, 170 km south of Anchorage. This predominately spruce stand contained trees that were ≈150 yr old with a mean dbh of 36 cm, a mean height of 30 m, and a stand density of 481 stems per ha. Ground cover was heavy and consisted mostly of devil's club, *Echinopanax horridum* (Sm. Dece. and Planch), with lesser amounts of alder, *Alnus* spp, and willow, *Salix* spp.

1999 Trapping Experiment. Twelve-unit, multiple funnel traps (Lindgren 1983) were hung from branches of nonhost or dead spruce trees, or on nylon rope suspended between host trees. Traps were hung at least 10 m apart (Bakke et al. 1983) with collection containers 0.3 m aboveground. Traps were baited with commercially available semiochemicals (PheroTech, Inc., Delta, BC, Canada) dispensed from either polyethylene bubble cap release devices or Eppendorf vials. The proto-type release devices containing MCH were obtained from Med-e-Cell, San Diego, CA (Table 1). This experimental dispenser, measuring 8.8 × 8.8 cm, consists of a battery-operated pumping module, a pheromone solution storage reservoir, and an absorbing collector pad. The collection pad, which absorbs the pheromone solution released by the micropump, allows for a high fluid evaporation rate, achieved by rapidly dispersing the solution over a large area of the pad. In this manner, the rate of pheromone evaporation into the environment is controlled by the micropump, rather than by diffusion or evaporation. Spruce beetles were collected from traps weekly from late May through July, the peak flight period for spruce beetles. Trapped insects were placed in labeled plastic bags and frozen for later identification and counting.

Table 1. Release rates of synthetic semiochemicals used in *D. rufipennis* studies, Alaska, 1999-2000

Semiochemical[a]	Releaser load (mg)	Release rate (mg/day)	Dispenser
Frontalin	300	2.6[b]	Eppendorf vial
alpha-pinene	600	1.5[b]	Eppendorf vial
MCOL	200	2.0[b]	Bubble cap
MCH/1999	440	5.0	Med-e-Cell
MCH/2000	440	2.6	Med-e-Cell
MCH	388	4.0[c]	Bubble cap

[a] Chemical purity > 98%.
[b] Release rates determined at 22°C.
[c] Release rates determined at 17°C.

Treatments were completely randomized and replicated six times. Treatments were: 1) attractant (*alpha*-pinene, frontalin, and 1-methyl-2-cyclohexen-1-ol [MCOL]); 2) attractant + one bubble cap of MCH; 3) attractant + MCH in Med-e-Cell releaser; and 4) unbaited traps as controls.

2000 Tree Protection Experiment. Twenty-one one-fifths ha plots (three treatments × 7 replications) were established 83 m apart in April 2000 near the Snow River. Treatments were randomly assigned and consisted of: 1) one-fifths ha (45 × 45 m) square untreated plot, 2) one-fifths ha square plot treated with 25 Med-e-Cell release devices; five releasers placed at 9 m intervals along five transects 9 m apart, 3) one-fifths ha round plot (25.5 m radius) treated with 25 Med-e-Cell release devices; 16 releasers placed along the outer perimeter at a 6 m spacing and nine releasers placed 6 m apart around an inner circle with a radius of 13.5 m. A smaller Med-e-Cell releaser was developed for the 2000 field study. The release rate of the Med-e-Cell releasers used in 2000 was less than the releasers used in the 1999 funnel trap study (Table 1). Spruce beetle pressure was low (<1 attacked tree per ha) in the study area. One 3-component spruce beetle attractant was placed on a dead spruce or nonhost near the center of each plot to ensure adequate beetle pressure throughout the study site.

After spruce beetle flight terminated in August, all trees in each plot were examined for evidence of spruce beetle attack.

Statistical Analyses. Statistical analyses were completed using "Statistix 7" software (Analytical Soft-

Table 2. Effect of MCH in bubble cap and Med-e-Cell releasers on the response of *D. rufipennis* to an attractant composed of frontalin, *alpha*-pinene, and MCOL (attractant) in multiple funnel traps, Anchorage, Alaska, 1999

Treatment	Spruce beetles (mean no./trap[a])
Attractant	381.0 ± 184.3a
Attractant + bubble cap	19.8 ± 10.2b
Attractant + Med-e-Cell	21.2 ± 10.8b
Unbaited trap	41.8 ± 20.4b

[a] Mean and standard error values followed by the same letter are not significantly different, $P > 0.05$, Tukey's comparison of means test.

ware, Tallahassee, FL). Numbers of *D. rufipennis* caught by each treatment and numbers of trees attacked by spruce beetle were first examined by the Shapiro-Wilk Test to determine whether data conformed to a normal distribution. Because they did not, data were transformed using the natural log + 1 before being subjected to analysis of variance (ANOVA) followed by Tukey's (1953) comparison of means test, $\alpha = 0.05$. Nontransformed means are reported in the results.

Results and Discussion

The addition of either MCH release device to the attractant blend significantly and equally reduced trap catches by ≈95% ($F = 6.95$, df = 3, 20, $P = 0.002$) (Table 2).

The mean number of previously-attacked trees ranged from 1.6 ± 0.7 per plot in the Med-e-Cell circular plots to 2.7 ± 1.5 in the square control plots, and the mean dbh of spruce ranged from 32.5 ± 1.5 cm in the Med-e-Cell square plots to 39.1 ± 3.1 cm in the Med-e-Cell circular plots. In neither case were these respective differences significant ($F = 0.5$, df = 2, 18, $P = 0.6$; $F = 3.0$, df = 2, 18, $P = 0.07$). The deployment of MCH in circular and square plots significantly reduced the number of new spruce beetle attacks by 79% and 87%, respectively ($F = 6.89$, df = 2, 18, $P = 0.006$) (Table 3). The deployment of MCH in round or square plots was not significantly different.

The significant reduction in the number of captured spruce beetles caused by MCH, regardless of the release device, confirms the results of previous studies (Holsten 1994, Zogas 2001). The success of MCH in Med-e-Cell devices in reducing the number of newly infested standing spruce in an area of low spruce beetle population density was the first such result in Alaska. Inconsistent release rates associated with passive releasers, such as bubble caps (Holsten et al. 2001), appear to have been eliminated by the use of the Med-e-Cell releaser. As there was no significant difference between the deployment of MCH in concentric circles or on a grid, future field studies should use square plots as they are easier to install and evaluate. Future studies should determine the minimum dosage required to achieve a significant treatment effect with differing spruce beetle population densities.

Table 3. Effect of MCH in Med-e-Cell releasers in 1/5 ha plots on the number of newly attacked spruce by *D. rufipennis*, Kenai Peninsula, Alaska, 2000

Treatment	Mean no. of newly attacked spruce/plot[a]
Check Plot	5.4 ± 2.1a
Med-e-Cell/Circular	1.1 ± 0.7b
Med-e-Cell/Square	0.7 ± 0.4b

[a] Mean and standard error values followed by same letter are not significantly different, $P > 0.05$, Tukey's comparison of means test.

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